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Toxoplasmosis in a Bobcat (*Felis rufus*)

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ABSTRACT: A bobcat (*Felis rufus*) estimated to be 6-mo old exhibited head pressing, stupor, intermittent seizures, and vocalization. Based on gross and histopathologic features, it was diagnosed to have severe focally extensive protozoal meningoencephalitis. *Toxoplasma gondii* was confirmed as the etiologic agent by an avidin-biotin immunohistochemical test. This is the first report of clinical toxoplasmosis in a free-ranging bobcat.

Key words: *Toxoplasma gondii*, toxoplasmosis, bobcat, *Felis rufus*, encephalitis, Georgia.

On 8 February 1993, a live female bobcat (*Felis rufus*) was submitted to the diagnostic laboratory of the Southeastern Cooperative Wildlife Disease Study exhibiting head pressing, stupor, intermittent seizures, and vocalization. The cat had been treed by dogs near a private residence in Irwin County (31°39′N, 83°23′W), Georgia (USA), on 7 February. The cat did not make further attempts to evade dogs or humans and was captured with a net. After a short period of observation, the cat was anesthetized with an intramuscular injection of 96 mg ketamine hydrochloride (Ketaset®, Fort Dodge Laboratories, Inc., Fort Dodge, Iowa, USA) and 3.8 mg xylazine (Rompun®, Mobay Corporation, Animal Health Division, Shawnee, Kansas, USA), and then euthanized with an intracardiac injection of 2 ml of euthanasia solution (Fatal-Plus®, Vortech Pharmaceuticals, Dearborn, Michigan, USA).

The cat was estimated to be 6-mo old based on deciduous canine tooth replacement (Crowe, 1975). It weighed 2.7 kg and was severely emaciated. The stomach was filled with a mass of hair containing two strips of material resembling sausage casings, each approximately 8 by 5 cm. A dark, fine granular exudate was observed on the meninges over a 23 mm diameter area of the dorsolateral aspect of the left cerebral hemisphere. A well demarcated, roughly circular area of pallor subjacent to this exudate and extending up to 14 mm into the neuropil was observed on gross examination following fixation in 10% neutral buffered formalin (Fig. 1).

At necropsy, blood samples were collected in tubes with and without ethylenediaminetetraacetic acid (EDTA) (Vacutainer®, Becton Dickinson Vacutainer Systems, Franklin Lakes, New Jersey, USA) and refrigerated at 4 C. Serum was separated from clotted blood and stored at −20 C. Portions of brain, liver, spleen, and small intestine were collected and frozen at −20 C. Portions of brain, trachea, lung, liver, spleen, kidney, adrenal gland, esophagus, stomach, small intestine, large intestine, tongue, diaphragm, and heart were fixed in 10% buffered formalin, embedded in paraffin, sectioned at 5 μm, and stained with hematoxylin and eosin. Unstained sections of paraffin-embedded brain were placed on Poly-L-Lysine® (Sigma Chemical Company, St. Louis, Missouri, USA) coated slides for immunohistochemical tests.

On microscopic examination, the brain had a focally extensive area of necrosis and inflammation of the leptomeninges, cerebral cortex, and subcortical white matter, particularly at the level of the hippocampus. Affected meninges were diffusely infiltrated by numerous plasma cells and lymphocytes with intense focal neutrophilic and histiocytic infiltration. Subjacent cerebral cortex was necrotic and diffusely infiltrated by numerous plasma cells, lymphocytes, neutrophils, macrophages, and gitter cells. Multifocal gliosis and numerous gemistocytes also were present. The major vascular lesion was cuffing of vessels by neutrophils, plasma cells, and lympho-

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cytes, but mild vasculitis also was present. Protozoal tachyzoites were seen primarily in phagocytic cells, but also were present in neurons, astrocytes, and free in the neuropil (Fig. 2). Microabscesses were scattered throughout areas adjacent to the large necrotic focus (Fig. 2). Inflammatory infiltrates extended deeply into subcortical white matter, frequently along blood vessels. There were no lesions in the cerebellum or pons.

In the lung there was moderate multifocal to coalescing, nonsuppurative interstitial pneumonia with occasional granulomatous foci containing protozoal tachyzoites. There was mild multifocal necrosis with scanty infiltrates of mononuclear leukocytes and neutrophils in the liver, renal pelvis, myocardium, and skeletal muscle of tongue and diaphragm. Tachyzoites were observed in hepatic and myocardial lesions. The lumen of the small intestine contained unidentified nematodes. There were no lesions in spleen, adrenal gland, trachea, esophagus, and stomach. Histologic lesions suggestive of canine distemper were absent.

Protozoa in brain sections were identified at the Zoonotic Diseases Laboratory, United States Department of Agriculture, Beltsville, Maryland (USA), with an avidin-biotin immunohistochemical procedure (Lindsay and Dubey, 1989). A microscopic agglutination test for antibodies against *Toxoplasma gondii* (Dubey and Desmonts, 1987) was performed on serum at the Beltsville Laboratory.

A portion of frozen brain was submitted to the Georgia Department of Human Resources, Atlanta, Georgia, for fluorescent antibody testing for rabies virus (Velleca and Forrester, 1981). Frozen brain, frozen small intestine, and refrigerated whole blood in EDTA were submitted to the Athens Diagnostic Laboratory, College of Veterinary Medicine, The University of Georgia, Athens, Georgia, for fluorescent antibody testing for feline infectious peritonitis virus, feline panleukopenia virus, and feline leukemia virus, respectively.

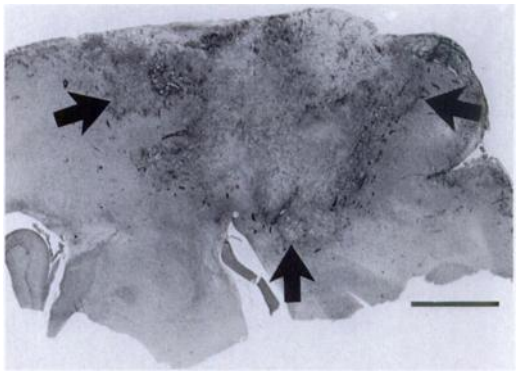


FIGURE 1. Area of necrosis and inflammation in the left cerebral hemisphere (arrows at periphery). H&E. Bar = 5 mm

Conjugates for these tests were obtained from American Bioresearch, Inc., Seymour, Tennessee (USA), and procedures followed Bedell et al. (1968). Formalin-fixed brain and lung were submitted to the James A. Baker Institute for Animal Health, College of Veterinary Medicine, Cornell University, Ithaca, New York (USA), for examination for canine distemper virus

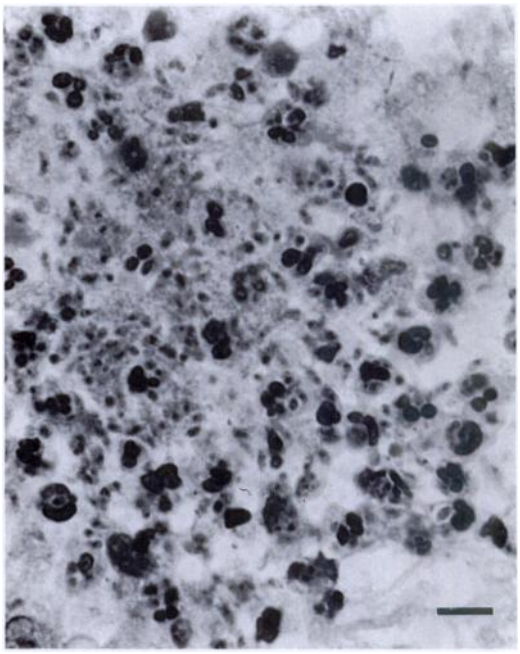


FIGURE 2. Microabscess with numerous *T. gondii* tachyzoites in neuropil. H&E. Bar = 10 μm.

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antigen by immunohistochemistry, as described by Appel et al. (1994).

Protozoa in brain sections reacted specifically with anti-*T. gondii* serum in the avidin-biotin immunohistochemical procedure. Based on the microscopic agglutination test, there was a serum antibody titer of 1:40,960 against *T. gondii*. Tests for rabies virus, feline infectious peritonitis virus, feline panleukopenia virus, feline leukemia virus, and canine distemper virus were negative.

Clinical toxoplasmosis is seen rarely in domestic or wild Felidae, even though infection with *T. gondii* is common (Dubey and Beattie, 1988). Felids, including bobcats, are the definitive hosts for *T. gondii*, as they are the only hosts capable of shedding oocysts (Miller et al., 1972). Dubey et al. (1987) reported fatal toxoplasmosis in a captive neonatal bobcat which apparently had been infected congenitally. Ours is the first report of clinical toxoplasmosis in a free-ranging bobcat.

Little information exists concerning the effect of concurrent infections on *T. gondii* infections in domestic cats. We found no evidence of the presence of a concurrent infection which could have predisposed this bobcat to toxoplasmosis, including canine distemper, an emerging problem in some wild Felidae (Appel et al., 1994). In experimental infections, young domestic cats develop clinical signs of toxoplasmosis following ingestion of tissue cysts more often than do adults (Dubey and Beattie, 1988). Therefore, age may have been a factor in this case. Despite this case, there is no additional evidence that toxoplasmosis constitutes a significant mortality factor for bobcats.

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